

Substitute Sequence Listing.TXT

<110> Korea Advanced Institute of Science and Technology
 <120> CONSTRUCTION OF NOVEL STRAINS CONTAINING MINIMIZING
 GENOME BY Tn5-COUPLED Cre/loxP EXCISION SYSTEM
 <130> 02730.0020.PCUS00
 <140> 10/505,328
 <150> PCT/KR02/02033
 <151> 2002-10-31
 <150> KR 10-2002-0009647
 <151> 2002-02-22
 <160> 13
 <170> KopatentIn 1.71
 <210> 1
 <211> 2437
 <212> DNA
 <213> Artificial Sequence
 <220>
 <223> TnKG1oxP

<400> 1	
attcaggctg cgcaactgtt gggaaggcgg atcgggtcgg gcctcttcgc tattacgcca	60
gctgtctctt atacacatct caaccatcat cgatgaattc gagctcggta cccgggttga	120
actgcggatc ttgcggccgc aaaaattaaa aatgaagttt tgacggatc gaacccaga	180
gtcccgtca gaagaactcg tcaagaaggc gatagaaggc gatgcgctgc gaatcgggag	240
cggcgatacc gtaaagcacg aggaagcggc cagcccattc gccgccaagc tcttcagcaa	300
tatcacgggt agccaacgct atgtcctgat agcggtcgc cacaccagc cgccacagt	360
cgatgaatcc agaaaagcgg ccattttcca ccgatgatatt cggcaagcag gcatcgccat	420
gggtcacgac gagatcctcg ccgtcgggca tccgcgcctt gagcctggcg aacagttcgg	480
ctggcgcgag cccctgatgc tcttcgtcca gatcatcctg atcgacaaga ccggcttcca	540
tccgagtacg tgctcgctcg atgcgatgtt tcgcttggtg gtcgaatggg caggtagccg	600
gatcaagcgt atgcagccgc cgcattgcat cagccatgat ggatacttcc tcggcaggag	660
caagggtgaga tgacaggaga tcttgccccg gcacttcgcc caatagcagc cagtccttc	720
ccgcttcagt gacaacgtcg agcacagctg cgcaaggaac gcccgctcgt gccagccagc	780
atagccgcgc tgccctcgtc tggagttcat tcagggcacc ggacaggctg gtcttgacaa	840
aaagaaccgg gcgccctcgc gctgacagcc ggaacacggc ggcatacagc cagccgattg	900
tctgtttgtc ccagtcatac ccgaatatgc tctccacca agcggccgga gaacctgcgt	960

Substitute Sequence Listing.TXT

gcaatccatc ttgttcaatc atgcgaaacg atcctcatcc tgtctcttga tccactagat	1020
tattgaagca tttatcaggg ttattgtctc atgagcggat acatatattga atgtatttag	1080
aaaaataaac aaataggggt tccgcgcaca ttccccgaa aagtgccacc tgcacatgat	1140
aattgatccg aagttcctat tctctagaaa gtataggaac ttcaaatgtt cgacaagctt	1200
gatctggtct atcgaaatta atacgactca ctatagggag accggaattc attatttgta	1260
gagctcatcc atgccatgtg taatcccagc agcagttaca aactcaagaa ggaccatgtg	1320
gtcacgcttt tcgttgggat ctttcgaaag ggcagattgt gtcacagagt aatggtgtgc	1380
tggtaaaagg acagggccat cgccaattgg agtattttgt tgataatggt ctgctagtgtg	1440
aacggatcca tcttcaatgt tgtggcgaat ttgaagtta gctttgattc cattcttttg	1500
tttgtctgac gtgatgtata cattgtgtga gttatagtgt tactcgagtt tgtgtccgag	1560
aatgtttcca tcttctttaa aatcaatacc ttttaactcg atacgattaa caagggtatc	1620
accttcaaac ttgacttcag cagcgtcttt gtagttcccg tcacttttga aagatatagt	1680
gcgttctctg acataacctt cgggcatggc actcttgaaa aagtcatgcc gtttcatatg	1740
atccggataa cgggaaaagc attgaacacc ataagagaaa gtagtgacaa gtgttgacca	1800
tggaacaggt agttttccag tagtgcaaat aaatttaagg gtaagttttc cgtatgttgc	1860
atcaccttca cctctccac tgacagaaaa ttgtgacca ttaacatcac catctaattc	1920
aacaagaatt gggacaactc cagtgaaga ttcttctctt ttactcattt tttctaccgg	1980
taccggggga tctctagag tcgacctgca ggcattgcaag ctggcgtaa tcatgttcat	2040
agctgtttcc tgtgtgaaat tgttatccgc tcacaattcc acacaacata cgagccggaa	2100
gcataaagtg taaagcctgg ggtgcctaag gagtgagcta actcacatta attgcgttgc	2160
gctcactgcc cgctttccag tcgggaaatc caaggcgcaa ttgcagctcg gtaccgggcc	2220
ccccctcgag ggacctata acttcgtata gcatacatta tacgaagtta tattaagggt	2280
tccggatcct ctagagtaga cctctagagt cgacctgcag gcatgcaagc ttcagggttg	2340
agatgtgtat aagagacagc tgcattaatg aatcggccaa cgcgcgggga gaggcggttt	2400
gcgtattggg cgctcttcgc cttcctcgct cactgac	2437

<210> 2
 <211> 1511
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> TnCloxP

<400> 2
 attcaggctg cgcaactgtt gggaagggcg atcgtgctcg gcctcttcgc tattacgcca 60

Substitute Sequence Listing.TXT

gctgtctctt atacacatct caaccatcat cgatgaattc gagctcggta ccgcaaaaat	120
taaaaatgaa gttttaaatc aatctaaagt atatatgagt aaacttggtc tgacaggttac	180
caatgcttaa tcagtgaggc accaataact gccttaaaaa aattacgccc cgccctgcc	240
ctcatcgag tactgttgta attcattaag cattctgccg acatggaagc catcacagac	300
ggcatgatga acctgaatcg ccagcggcat cagcaccttg tcgccttgcg tataatat	360
gcccatggtg aaaccgggg cgaagaagtt gtccatattg gccacgttta aatcaaaaact	420
ggtgaaactc acccagggat tggctgagac gaaaaacata ttctcaataa accctttagg	480
gaaataggcc aggtttttcac cgtaacacgc cacatcttgc gaatatatgt gtagaaactg	540
ccggaatcg tcgtggtatt cactccagag cgatgaaaac gtttcagttt gctcatgga	600
aacggtgtaa caagggtgaa cactatccca tatcaccagc tcacgctctt tcattgccat	660
acggaatttc ggatgagcat tcactcaggc ggcaagaatg tgaataaagg ccggaataaa	720
ctgtgcttta tttttcttta cggcttttaa aaaggccgta atatccagct gaacggctcg	780
gttataggtg cattgagcaa ctgactgaaa tgcccaaaa tgttctttac gatgccattg	840
ggatatatca acggtggtat atccagtgat tttttctcc attttagctt ccttagctcc	900
tgaaaatctc gataactcaa aaaatacgcc cggtagtgat cttatttcat tatggtgaaa	960
gttggaacct cttacgtgcc gatcaacgct tcattttcgc caaaagtgg cccagggtt	1020
cccggtatca acagggacac caggatttat ttattctgct aagtgtatct ccgtcacagg	1080
tatttattcg gcgcaaatg cgtcgggtga tgctgccaac ttactgattt agtgtatgat	1140
ggtgtttttg aggtgctcca gtggcttctg tttctatcag catcgatgaa ttgatccgaa	1200
gttcctattc tctagaaagt ataggaactt cgaattgtcg acaagcttga tctggcttat	1260
cgaattaat acgactcact atagggagac cggaattcga gctcggtagc gggccccccc	1320
tcgagggacc taataacttc gtatagcata cattatacga agttatatta agatcctcta	1380
gagtcgacct gcaggcatgc aagcttcagg gttgagatgt gtataagaga cagctgcatt	1440
aatgaatcgg ccaacgcgcg gggagaggcg gtttgcgat tgggcgctct tccgcttcct	1500
cgtcactga c	1511

<210> 3
 <211> 19
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> OE sequence

<400> 3
 ctgtctctta tacacatct

19

Substitute Sequence Listing.TXT

<210> 4
<211> 34
<212> DNA
<213> Artificial Sequence

<220>
<223> loxP site

<400> 4
ataacttcgt atagcatata ttatacgaag ttat

34

<210> 5
<211> 996
<212> DNA
<213> Artificial Sequence

<220>
<223> KmR gene

<400> 5
gcaaaaatta aaaatgaagt tttagcggtg tcgaacccca gagtcccgtc cagaagaact 60
cgtaagaag gcgatagaag gcgatgcgt gcgaatcggg agcggcgata ccgtaaagca 120
cgaggaagcg gtcagcccat tcgccgcaa gctcttcagc aatatcagg gttagccaag 180
ctatgtcctg atagcggtcc gccacacca gccggccaca gtcgatgaat ccagaaaagc 240
ggccattttc caccatgata ttgcgaagc aggcacgccc atgggtcacg acgagatcct 300
cgccgtcggg catccgcgcc ttgagcctgg cgaacagttc ggctggcgcg agcccctgat 360
gctcttcgtc cagatcatcc tgatcgaca gaccggcttc catccgagta cgtgctcgtc 420
cgatgcgatg tttagccttg tggcgaatg ggcaggtagc cggatcaagc gtatgcagcc 480
gccgcattgc atcagccatg atggatactt tctcggcagg agcaaggtag gatgacagga 540
gatcctgccc cggcacttcg cccaatagca gccagtcctt tcccgttcga gtgacaacgt 600
cgagcacagc tgcgcaagga acgcccgtcg tggccagcca cgatagccgc gctgcctcgt 660
cttggagttc attcagggca ccggacaggt cggctttgac aaaaagaacc gggcgcccct 720
gcgctgacag ccggaacacg gcggcatcag agcagccgat tgtctgtgtg gccagtcac 780
agccgaatg cctctccacc caagcggcgg gagaacctgc gtgcaatcca tctgttccaa 840
tcattgcgaa cgatcctcat cctgtctctt gatccactag attattgaag catttatcag 900
ggttattgtc tcattgagcg atacatattt gaattgtattt agaaaaataa acaaataggg 960
gttccgcgca catttccccg aaaagtgcga cctgca 996

<210> 6
<211> 947
<212> DNA
<213> Artificial Sequence

Substitute Sequence Listing.TXT

<220>

<223> GFP gene

<400>

6

attatttcta	gagctcatcc	atgccatgtg	taatcccagc	agcagttaca	aactcaagaa	60
ggaccatgtg	gtcacgcttt	tcgttgggat	ctttcgaaag	ggcagattgt	gtcgacagggt	120
aatggtttgc	tggtaaaagg	acagggccat	cgccaattgg	agtattttgt	tgataatgggt	180
ctgctagtgt	aacggatcca	tcttcaatgt	tgtggcgaat	tttgaagtta	gctttgattc	240
cattcttttg	ttgtctgccc	gtgatgtata	cattgtgtga	gttatagttg	tactcgagtt	300
tgtgtccgag	aatgtttcca	tcttctttta	aatcaatacc	ttttaactcg	atacgattaa	360
caaggggtatc	accttcaaac	ttgacttcag	cacgcgtctt	gtagttcccg	tcattcttga	420
aagatatagt	gcgttctctg	acataacctt	cgggcatggc	actcttgaaa	aagtcatgcc	480
gtttcatatg	atccggataa	cgggaaaagc	attgaacacc	ataagagaaa	gtagtgcaca	540
gtgttggcca	tggaacagggt	agttttccag	tagtgcaaat	aaatttaagg	gtaagttttc	600
cgtatgttgc	atcaccttca	ccctctccac	tgacagaaaa	tttgtgccca	ttaacatcac	660
catctaattc	aacaagaatt	gggacaaact	cagtgaaaag	ttcttctcct	ttactcattt	720
tttctaccgg	taccggggga	tcctctagag	tcgacctgca	ggcatgcaag	cttggcgtaa	780
tcattggtcat	agctgtttcc	tgtgtgaaat	tgttatccgc	tcacaattcc	acacaacata	840
cgagccggaa	gcataaagtg	taaagcctgg	ggcgccaat	gagtgagcta	actcacatta	900
attgcgttgc	gctcactgcc	cgctttccag	tcgggaaatc	caaggggc		947

<210>

7

<211> 1069

<212> DNA

<213> Artificial Sequence

<220>

<223> CMR gene

<400>

7

gcaaaaaatta	aaaatgaagt	tttaaatcaa	tctaaagtat	atatagtagta	acttggcttg	60
acagttacca	atgcttaatc	agtgaggcac	caataactgc	cttaaaaaaa	ttacgccccg	120
ccttgccact	catcgagcta	ctgttgtaat	tcattaagca	ttctgccgac	atggaagcca	180
tcacagacgg	catgatgaac	ctgaatcgcc	agcggcatca	gcaccttgtc	gccttgcgta	240
taatatattgc	ccatgggtgaa	aacgggggcg	aagaagttgt	ccatattggc	cacgtttaaa	300
tcaaaactgg	tgaaactcac	ccaggggattg	gctgagacga	aaaacatatt	ctcaataaac	360
cctttaggga	aataggccag	gttttcaccg	taacacgcca	catcttgcca	atatatgtgt	420

Substitute Sequence Listing.TXT

agaaactgcc	ggaaatcgtc	gtgggtattca	ctccagagcgc	atgaaaacgt	ttcagtttgc	480
tcatggaaaa	cggtgtaaca	agggtgaaca	ctatcccata	tcaccagctc	accgtctttc	540
attgccatac	ggaatttcgc	atgagcattc	atcaggcggg	caagaatgtg	aataaaggcc	600
ggataaaact	tgtgcttatt	tttctttacg	gtctttaaaa	aggccgtaat	atccagctga	660
acggtctggt	tataggtaca	ttgagcaact	gactgaaatg	cctcaaaatg	ttctttacga	720
tgccattggg	atatatcaac	ggtggtatat	ccagtgattt	ttttctccat	tttagcttcc	780
ttagctcctg	aaaaatctga	taactcaaaa	aatacgcccgc	gtagtgatct	tatttcatta	840
tggtgaaagt	tggaacctct	tacgtgccga	tcaacgtctc	attttcgccca	aaagtgggcc	900
cagggtcttc	cggtatcaac	agggacacca	ggattttatt	attctgcgaa	gtgatcttcc	960
gtcacaggta	ttttatcggc	gcaaaagtgcg	tcgggtgatg	ctgcccaact	actgatttag	1020
tgtatgatgg	tggtttttgag	gtgctccagt	ggcttctggt	tctatcagc		1069

<210> 8
 <211> 19
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> primer-pMOD<MCS>FP-1

<400> 8
 attcaggctg cgcaactgt 19

<210> 9
 <211> 22
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> primer-pMOD<MCS>RP-1

<400> 9
 tcagtgagcg aggaagcgga ag 22

<210> 10
 <211> 28
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> primer-Tn5Ext

<400> 10
 agcatacatt atacgaagtt atattaag 28

<210> 11

Substitute Sequence Listing.TXT

<211>	35	
<212>	DNA	
<213>	Artificial Sequence	
<220>		
<223>	primer-Arb1	
<220>		
<221>	misc_feature	
<222>	(21)..(30)	
<223>	n is a or g or c or t	
<400>	11	
	ttgagcgata gacgtacgat nnnnnnnnnn gatat	35
<210>	12	
<211>	20	
<212>	DNA	
<213>	Artificial Sequence	
<220>		
<223>	primer-Arb2	
<400>	12	
	ttgagcgata gacgtacgat	20
<210>	13	
<211>	25	
<212>	DNA	
<213>	Artificial Sequence	
<220>		
<223>	primer-Tn5Int	
<400>	13	
	tcgacctgca ggcattgcaag ctctca	25